NAME: Turner Designs10-AU-30 Fluorometer S/N 1100209

1) Introduction

The Turner 10-AU-30 fluorometer is equipped with a 10-040R optical kit that includes the following:

- a. Light Source: 10-089, Blue Lamp
- b. Reference Filter 10-032, Filter: 400-700nm Square 1
- c. Excitation Filter: 034-0392, Filter-436 FS10-25
- d. Emission Filter: 034-0395, Filter-680 FS10-25

2) Calibration/Maintenance

The protocol for calibration followed that which is published in the Turner designs manual. Briefly, the sensitivity was adjusted first based on a chosen single calibration point that represented the medium concentration range of expected Chlorophyll *a* concentrations. Next, the instrument was calibrated using the aforementioned standard as a single point. The concentration of this standard represented approximately 80% of the highest expected concentration of Chlorophyll *a*. A linear calibration curve consisting of a total of 7 points was developed around the single point standard. The calibration is routinely monitored using chlorophyll a solutions or a solid standard.

3) Sample Extraction and Measurement

A. Extraction:

- i. A known volume of water was filtered onto a 25mm GF/F filter. Samples were collected in triplicate.
- ii. The filter was transferred to a 15ml plastic Eppendorf tube to which 7ml of 90% acetone was added.
- iii. The samples was agitated and placed in a -20°C freezer for 24 hours.

B. Measurement:

- i. The fluorometer was allowed to warm up for at least 15 minutes prior to use.
- ii. Calibration stability was monitored using a Turner solid standard (P/N 10-AU-904).
- iii. After allowing the samples to warm to room temperature in the dark, each sample was transferred to a 13x100mm glass cuvette (VWR 47729-572).
- iv. Raw fluorescence was read and recorded.

4) Data processing

The total Chlorophyll a concentration of each sample was calculated using the slope and y intercept from the linear calibration curve (x=y-b/m).

5) Data Reporting Mean and standard deviation of CHL replicates will be reported.

6) Reporting Notation

CHL: Fluorometrically-derived Chlorophyll *a* CHL_sd: Standard deviation of replicates